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Abstract: We describe cases of collyriclosis in apodiform and passeriform birds in Portugal, Switzerland, and Germany. We extend the host range of Collyriculm faba to include apodiform birds (Apus apus, Apus melba, and Apus pallidus) and the passerine Sitta europaea (Eurasian Nuthatch). Infections varied in severity from an incidental finding to severe debilitation and death. The infection route remains unclear with the apparent absence from Germany, Portugal, and Switzerland of the first intermediate host of C. faba, the aquatic gastropod Bythinella austriaca, implying that other organisms might be involved in the parasite's life cycle. Furthermore, the detection of C. faba cysts in very young passerine birds may indicate an infection during the nestling stage and a rapid development of parasite containing subcutaneous cysts. This series of cases highlights an increased geographic range into Portugal and the potential debilitating nature of a parasite of migratory birds in Europe. However, given the rarity of cases, collyriclosis does not seem to present an important threat to migratory species preservation.

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Stamatios A. Tahas,[1,12] Anastasia Diakou,[2] Monika Dressel,[3] Samuel Frei,[4] Fábia M. Pinto Azevedo,[5] María V. Mena Casero,[5] Carla Maia,[6,7] Paula Grest,[8] Felix Grimm,[9] Jiljí Sitko,[10] and Ivan Literák[11] [1]Clinic for Zoo Animals, Exotic Pets and Wildlife, Vetsuisse Faculty, University of Zurich, Winterthurerstrasse 260, 8057 Zurich, Switzerland; [2]Laboratory of Parasitology and Parasitic Diseases, School of Veterinary Medicine, Faculty of Life Sciences, Aristotle University of Thessaloniki, 54124 Thessaloniki, Greece; [3]Dressel Small Animal Practice, Bahnhofstrasse 3, 79585 Steinen, Germany; [4]Wuppertal Zoo, Hubertusallee 30, 42117 Wuppertal, Germany; [5]Wildlife Rehabilitation and Investigation Center - RIAS, Apartado 1009, 8700-282 Olhão, Portugal; [6]Global Health and Tropical Medicine, Medical Parasitology Unit, Institute of Hygiene and Tropical Medicine, New University of Lisbon, Rua de Jungueira 100, 1249-008 Lisbon, Portugal; [7]Faculty of Veterinary Medicine, University Lusofona of Humanities and Technologies, Campo Grande, 376, 1749-024 Lisbon, Portugal; [8]Institute of Veterinary Pathology, Vetsuisse Faculty, University of Zurich, Winterthurerstrasse 268, 8057 Zurich, Switzerland; [9]Institute of Parasitology, Vetsuisse Faculty, University of Zurich, Winterthurerstrasse 266a, 8057, Zurich, Switzerland; [10]Comenius Museum, Moravian Ornithological Station, Horní nám. 1, 75152 Přerov, Czech Republic; [11]Department of Biology and Wildlife Diseases, Faculty of Veterinary Hygiene and Ecology, University of Veterinary and Pharmaceutical Sciences, Palackého tř. 1, 61242 Brno, Czech Republic; [12]Corresponding author (email: stahas@vetclinics.uzh.ch)

ABSTRACT: We describe cases of collyriclosis in apodiform and passeriform birds in Portugal, Switzerland, and Germany. We extend the host range of *Collyriculm faba* to include apodiform birds (*Apus apus, Apus melba*, and *Apus pallidus*) and the passerine *Sitta europaea* (Eurasian Nuthatch). Infections varied in severity from an incidental finding to severe debilitation and death. The infection route remains unclear with the apparent absence from Germany, Portugal, and Switzerland of the first intermediate host of *C. faba*, the aquatic gastropod *Bythinella austriaca*, implying that other organisms might be involved in the parasite's life cycle. Furthermore, the detection of *C. faba* cysts in very young passerine birds may indicate an infection during the nestling stage and a rapid development of parasite-containing subcutaneous cysts. This series of cases highlights an increased geographic range into Portugal and the potential debilitating nature of a parasite of migratory birds in Europe. However, given the rarity of cases, collyriclosis does not seem to present an important threat to migratory species preservation.

Key words: Apus apus, *Apus melba*, *Apus pallidus*, *Collyriclum faba, Passer domesticus*, pericloacal cysts, *Phoenicurus ochruros*, *Sitta europaea*.

Collyriclum faba is a trematode parasite predominantly of passerine birds that has been identified in Europe, America, Africa, and Asia (Stunkard 1971; Rzad and Busse 2015). It usually presents as subcutaneous pericloacal cysts, though other presentations are also reported (Literák and Sitko 2006). A small opening is usually visible on the top of cysts and paired semicircular parasites measuring 0.4×0.5×0.3 cm are identified on cyst dissection (Tyzzer 1918; Literák and Sitko 1997). The parasite's life cycle has only recently been elucidated, with the aquatic gastropod *Bythinella austriaca* as the first intermediate host, in which the formation of developmental stages occurs, with mayflies *Ecdyonurus venosus* and *Rhithrogena picteti*, as the second intermediate hosts (Heneberg et al. 2015).

In May 2011, a juvenile Eurasian Nuthatch (*Sitta europaea*) was presented to a local veterinary practice in Steinen, southern Germany (47°38′N, 7°44′E) due to a cat bite wound. Two hard subcutaneous pericloacal cystic structures measuring 0.4 cm in diameter were noted on physical examination. Histopathologic examination of the structures revealed multiple cysts filled with intraluminal trematodes. Cysts were associated with a granulomatous reaction containing mainly macrophages, lymphocytes, and plasma cells. Histopathologic examination identified *C. faba* infection (Tyzzer 1918; Blankespoor et al. 1985). Following surgery for cyst removal, the bird was successfully treated and released into the wild in July 2011.

Three additional cases of collyriclosis in passerine birds were seen in the same veterinary practice. A juvenile House Sparrow

FIGURE 1. Severe pericloacal collyriclosis in a Pallid Swift (*Apus pallidus*) from Portugal. There is severe edema and inflammation of the pericloacal tissues. The small hole from which the adult parasites release eggs is evident as a black dot (arrow).

(*Passer domesticus*) was presented in 2011 due to severe feather damage. Pericloacal and femoral cysts typical of *C. faba* were identified as previously described (Literák et al. 2003). The animal died despite treatment. A high coccidian burden was identified on fecal examination. In 2015, a House Sparrow and a Black Redstart (*Phoenicurus ochruros*), rescued as nestlings and hand reared, were presented when subcutaneous pericloacal cysts, identical with *C. faba* cysts, were observed around the time of fledging. Both birds were treated with doxycycline (Ratiopharm, Ulm, Germany), praziquantel (Selectavet, Weyarn-Holzolling, Germany), and ivermectin (Merial, Hallbergmoos, Germany). Following treatment, contents of the cysts were aspirated. Both birds recovered without complications and were released into the wild when fully fledged. No further cyst formation was noted.

Five adult Pallid Swifts (*Apus pallidus*) and one adult Alpine Swift (*Apus melba*) were presented to the Wildlife Rehabilitation and Investigation Center (RIAS) in Olhão, Portugal (37°01′N, 7°48′W). In April–May 2013, all birds were presented due to the inability to fly. Anomalous pink, oval cutaneous pericloacal cysts measuring 0.4 to 4 cm in diameter were noted on all birds (Fig. 1). One to five black spots were noted on the lesions. Due to severe inflammation and necrosis, four birds were euthanized following presentation, whereas two died despite supportive care. Postmortem examinations showed no lesions other than the cloacal cysts that involved most of the ventral coelomic wall. The cysts had characteristics typical of *C. faba* (Tyzzer 1918;

Literák et al. 2003). Parasites in cysts were morphologically identified as immature stages of *C. faba* (Tyzzer 1918; Literák et al. 2003). Following processing for identification, no sample material suitable for deposit as a voucher specimen in a museum collection remained.

An adult male Common Swift (*Apus apus*) was presented to the Clinic for Zoo Animals, Exotic Pets and Wildlife in Zurich, Switzerland (47°23′N, 8°33′E) in June 2015 due to trauma. Following radiography, the animal was euthanized due to an intraarticular scapular fracture. An incidental finding of three subcutaneous pericloacal cysts (0.4–0.7 cm) was noted, from which dark fluid was aspirated postmortem. Multiple oval-shaped trematode eggs measuring 19×11 µm were identified as eggs of *C. faba* (Tyzzer 1918; Grove et al. 2005). Postmortem evaluation of the Common Swift revealed no significant abnormalities. Cyst histopathology revealed a broad rim of granulation tissue that surrounded the parasite, with a mixed cellular inflammation consisting of lymphocytes, histiocytes, giant cells, and granulocytes. Within the parasite, the uterus was filled with numerous eggs with a yellow to brown shell (Fig. 2).

Genomic DNA was isolated from adult individuals by using a commercial kit according to the manufacturer's instructions (QIAamp DNA Mini Kit, Qiagen, Hilden, Germany) and fragments of the 18S ribosomal (r) RNA and the ITS2 genes (including flanking 5.8S and 28S rRNA) were amplified as previously described (Heneberg et al. 2015). Sequences showed identities of 100% (346/346 base pairs, 18S rRNA, GenBank accession no. JK231122) and >99% (588/589 base pairs, ITS2, GenBank accession no. JK231122) with sequences published for *C. faba*.

The parasite *C. faba* has not been reported from apodiform birds. It has previously been reported in a House Sparrow in Switzerland (Denzler and Lobsiger Molliet 1991) and in Black Redstarts in Europe but not in Portugal and not in Eurasian Nuthatches (Literák et al. 2003).

Despite a long history of caring for apodiform and passeriform birds, no previous case of collyriclosis has been noted by the Clinic for Zoo Animals, Exotic Pets and Wildlife in Switzerland or RIAS in Portugal. The rarity of cases reported could be due to suboptimal recording, death of infected birds before presentation, dismissing lesions as other possible etiologies, or most likely, the low prevalence of *C. faba* infection. The prevalence of *C. faba* in European hirundines, which feed on aerial insects similarly to swifts, has been reported to be one in 20,641 Sand Martins (*Delichon urbica*) and one in 4,484 Barn Swallows (*Hirundo rustica*) in Central Europe (Heneberg et al. 2011). Even in passerines, in which infection is more prevalent, it is reported to be rare in Europe (Denzler and Lobsiger Molliet 1991). The relatively frequent cases observed in southern Germany are likely to be due to the highly focal occurrence of collyriclosis in birds in Europe (Literák and Sitko 2006; Heneberg et al. 2015).

The time required for the development of subcutaneous cysts has been shown to be as short as 13–19 d in passerine birds (Literák et al. 2003) but has not been fully elucidated (Heneberg et al. 2015) and is not known in apodiform birds. In particular, the development of *C. faba* cysts in hand-raised birds allows for two possibilities of infection and

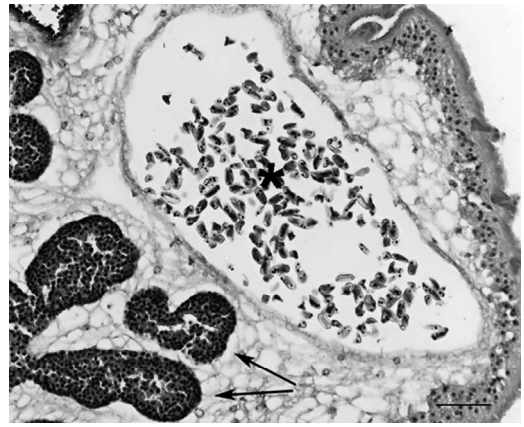

FIGURE 2. *Collyriclum faba* in a Common Swift (*Apus apus*) in Switzerland. Trematode body with vitellaria (arrows) and eggs (asterisk), lined by a tegument that contains spines. H&E staining. Bar=100 µm.

development of cysts. The first is that birds were infected by being fed mayflies, their nymphs or subimagos, or other invertebrates acting as intermediate hosts while still in the nest. This allows a period for the cysts to develop between birth and the fledgling stage of young birds, typically less than 5 wk in passerines (Blem 1975; Leedman and Magrath 2003; Draganoiu et al. 2006). The less likely possibility is infection during hand rearing, which would allow a shorter period for the cysts to become detectable, between nestling (when found) and fledgling (when cysts were noted), which in the two previously mentioned cases was less than 2 wk. However, the feeding of nestlings in this period with mealworm beetle (*Tenebrio molitor)* larvae and a commercial protein mix for young birds (Claus Nestlingsfutter, Claus, Germany) does not support the latter theory, given the current understanding of the parasite's life cycle (Heneberg et al. 2015).

The identification of *C. faba* cysts in subadult nonmigratory passerines in southern Germany, as well as earlier demonstrations of the parasites in nonmigratory European passerines (Literák et al. 2003) proves that a local development of the parasites occurs in Central Europe. Geographic analysis of the occurrence of *C. faba* cysts in migrating and resident birds has previously identified the Alps and their foothills as an endemic area (Literák and Sitko 2006).

However, the question of whether the swifts were infected before, during, or after migration remains unanswered. The lack of reports of subcutaneous collyriclosis in East Africa, whence swifts migrate to Switzerland and Portugal (Åkesson et al. 2012), the absence of the proposed first intermediate host (*B. austriaca*), and the scarcity of related snail species in Africa (Garcia et al. 2010), as well as our report that the parasites found in birds from Portugal were mostly immature stages adds weight to the suspicion that infection took place in Europe.

The likelihood that the infection of the previously mentioned birds occurred in Europe needs to be discussed with regard to the distribution of known or possible intermediate hosts. The aquatic gastropod *B. austriaca* that has been identified as a first intermediate host (Heneberg et al. 2015) has not been demonstrated in Switzerland (Fehér et al. 2010) and Germany with the exception of Bavaria, where its population is classified as endangered (Glöer and Meier-Brook 2003). The genus also seems to be absent from Portugal (Fehér et al. 2010). This allows speculation that other aquatic mollusks might act as primary intermediate hosts of *C. faba* or that yet unidentified focal populations of *B. austriaca* do indeed occur in these areas. This should be considered also for the possible infection in East Africa. A different *Bythinella* species is present in northern Africa but is classified as critically endangered (Garcia et al. 2010) so it is considered unlikely that it supports the *C. faba* life cycle. Alternative swift migration routes in which the parasite and intermediate hosts may be endemic, i.e., the Carpathian Mountains (Literák et al. 2003) may be considered but seem unlikely.

The clinical importance of the infection, especially in nature, remains unclear. Under human care, fatal cases are uncommon and are usually due to coinfections (Literák et al. 2003; Grove et al. 2005). The cases from Portugal demonstrate uncommon fatal cases, as no other obvious causes for mortality were noted. The Common Swift examined in Switzerland was in good general health, suggesting an incidental finding. The passerine cases from southern Germany remain the first cases of surgical or medical treatment of collyriclosis in truly wild birds that were subsequently successfully released. The positive curative outcome in the predominantly nonmigratory passerines in Germany, as opposed to a guarded prognosis in Alpine and Pallid Swifts, may be due to the migratory nature of the latter two species. It is hypothesized that migratory species may be immunocompromised and more susceptible to infections shortly after migration (Owen and Moore 2006; Buehler et al. 2008). Regarding species conservation, given the apparent rarity of collyriclosis, we consider it unlikely that the infection compromises survival of any of the species mentioned.

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